

Poster Presentation

LA.P37

Assembly of Sesbania mosaic virus: Role of the coat protein N-terminal R domain

A. Gulati¹, H. Savithri², M. Murthy¹

¹Indian Institute of Science, Molecular Biophysics Unit, Bangalore, India, ²Indian Institute of Science, Department of Biochemistry, Bangalore, India

Coat proteins of several isometric viruses consist of two domains, a disordered N-terminal R-domain consisting of several positively charged residues and a shell (S) domain characterized by a jelly roll β -barrel structure. The three-dimensional structure of Sesbania mosaic virus (SeMV), a T=3 plant virus, has been determined at 3 Å resolution. The full length coat protein, when expressed in E. coli, assembles into T=3 icosahedral shells (VLPs) resembling native virus particles. In the present investigations, the role of N-terminal R domain in the assembly of VLPs was explored by replacing the R domain with a presumably globular domain (SeMV-P10) and other intrinsically disordered (SeMV-P8, and SeMV-VPg) SeMV encoded domains. The R domain was also replaced with the non-viral globular B-domain of Staphylococcus aureus protein A. These domains were of nearly the same size as that of the R-domain. Most of the chimeric coat proteins, when expressed in E.coli, formed VLPs, which could be purified by ultra-centrifugation. The purified VLPs were examined by transmission electron microscopy (TEM), which suggested that a fraction of the expressed proteins could assemble into T=3 VLPs, although often, the particles were heterogeneous. Interestingly, the SeMV N Δ 65B CP could also be purified by Ni-NTA chromatography as a dimer which assembled into T=1 VLPs under crystallization conditions. The structure of N Δ 65B-CP VLPs revealed that the assembled particles were devoid of divalent metal ions at the canonical site and there was no density corresponding to the B domain. However, the S domain could be superimposed on that of SeMV N Δ 65VLPs determined earlier. The other VLPs-SeMVN Δ 65P10 CP, SeMVN Δ 65P8 CP and SeMVN Δ 65VPg could not be crystallized because of their heterogeneity. These studies suggest a subtle interplay between the length, sequence and structure of the R-domain polypeptide and the assembly of particles.

Keywords: Virus assembly, Disordered R-domain, Capsid protein